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**Title:** Quality of Fresh-Cut Apple Slices as Affected by Low-Dose Ionizing Radiation and Calcium Ascorbate Treatment

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# Quality of Fresh-cut Apple Slices as Affected by Low-dose Ionizing Radiation and Calcium Ascorbate Treatment

XUETONG FAN, BREDAN A. NIEMERA, JAMES P. MATTHEIS, HONG ZHUANG, AND DOUGLAS W. OLSON

**ABSTRACT:** Although ionizing radiation effectively inactivates food-borne bacterial pathogens in fresh-cut fruits and vegetables, it may adversely affect product quality. In this study, the effects of calcium ascorbate (CaA) and ionizing radiation on quality of 'Gala' apple slices under modified atmosphere packaging were investigated. 'Gala' apple slices, treated with water or 7% CaA followed by either nonirradiation (0 kGy) or irradiation at 0.5 and 1.0 kGy, were stored at 10 °C for up to 3 wk. The titratable acidity, pH, firmness, ascorbic acid content, color, and microflora population were measured weekly throughout storage. Irradiation did not affect titratable acidity and pH of sliced apples. Fruit slices softened during irradiation and storage, but this decrease in firmness during storage was reduced by the CaA treatment. Although the ascorbic acid content of apple slices treated with CaA decreased rapidly during storage, the ascorbic acid content was always higher in those treated samples than in the apple slices treated with water. Irradiation decreased both  $L^*$  and hue values of apple slices. Hue values decreased during the entire storage period while  $L^*$  increased during the 1st wk of storage, then decreased between 1 to 3 wk of storage. CaA increased  $L^*$  and hue values of apple slices, suggesting CaA reduced browning, even in irradiated samples. The microflora population of apples slices was not affected by CaA, and CaA treatment did not alter the reduction in microflora by irradiation. The combination of CaA and irradiation enhanced microbial food safety while maintaining quality of fresh-cut apple slices.

**Keywords:** irradiation, fresh-cut apple slices, calcium ascorbate, browning, firmness

## Introduction

Although the number of recalls and outbreaks is low, food-borne incidents associated with consumption of fresh produce have increased in the last decade (CDC 2000). Fresh-cut produce is of special concern because its consumption has been increasing 10% to 20% per year (IFPA 2004), and it is routinely consumed raw, without any further antimicrobial processing. To prolong shelf life and enhance safety, fresh-cut produce, including apples, is often stored in modified atmosphere packaging (MAP) and at refrigeration temperatures. However, *L. monocytogenes* can grow at low temperatures (4 to 6 °C) even under modified atmosphere (Jacxsens and others 1999). There was at least 1 recall associated with fresh-cut apples due to possible contamination with *L. monocytogenes* (FDA 2001). Therefore, technology is needed to inactivate food-borne pathogens in fresh-cut apples and other fresh-cut products.

Ionizing radiation is a nonthermal processing technology that effectively inactivates food-borne pathogens in many foods, including fresh-cut produce (Niemira and others 2003; Prakash and others 2000). Besides its effectiveness in eliminating food-borne pathogens, ionizing radiation also reduces spoilage microorganisms, inhibits ethylene production/action, and retards the ripening process, resulting in extended shelf-life (Thomas 1986).

However, ionizing radiation may have adverse effects on fresh

produce including fresh-cut apples. These effects may include fruit softening and decreased ascorbic acid (AA) and titratable acidity (TA). Gunes and others (2001) found irradiation at doses above 0.34 kGy reduced firmness of fresh-cut apples. The loss of AA has been observed in some fresh fruits and vegetables (Graham and Stevenson 1997). Whether irradiation reduces AA content in fresh-cut apples is not clear. The TA is a major factor influencing flavor of fresh-cut apples. Earlier studies have shown that TA of whole apples (Fan and Mattheis 2001) and in fresh-cut apples (Rocha and Morais 2003) decreased during storage. There are very few studies about irradiation effects on TA in fresh-cut apples.

Cutting of whole apples results in cellular delocalization of enzymes and their substrates, and exposure of the cut surface to oxygen, leading to enzymatic browning catalyzed by polyphenol oxidase. Enzymatic browning is a major factor limiting shelf-life of fresh-cut apples. Many antibrowning agents have been developed, such as sulfur dioxide, ascorbic acid and its derivatives, L-cysteine, 4-hexylresorcinol, and calcium (Sapers and others 2002). Combination of AA and  $\text{CaCl}_2$  inhibited tissue browning of fresh-cut apples (Ponting and others 1972) and pears (Gorny and others 1998) for several weeks.

Calcium ascorbate (CaA), which is one of the common antibrowning agents that is currently used by the fresh-cut apple industry (Chen and others 1999; Karaibrahimoglu and others 2004), also affects other properties of apple slices. Calcium prevented irradiation-induced softening in thin apple slices (3 to 4 mm thick), but was not effective with thicker wedges (Gunes and others 2001). Rocha and others (1998) found that treating apple slices with 0.75% AA, alone or in combination with 0.75%  $\text{CaCl}_2$ , inhibited not only enzymatic browning but also loss of firmness during storage. CaA can be used at concentrations up to 9%. Treating apple slices with CaA will prob-

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ably increase their AA levels, therefore compensating the AA loss due to irradiation, but this has not been documented. AA is a strong antioxidant, and the use of antioxidants may provide protection against adverse effects caused by irradiation. However, antioxidants may also increase radiation resistance of microorganisms as shown in solutions (Sharma and others 2000; Sommers and others 2002).

The objective of this study was to investigate the effects of CaA and irradiation on quality attributes and microflora of fresh-cut 'Gala' apples packaged in MAP during storage.

## Materials and Methods

### Fruit source and assessment of fruit maturity

'Gala' apple fruit (*Malus x domestica* Borkh.) were harvested at commercial maturity from 9-year-old trees in an orchard in Medford, N.J., U.S.A. The fruit were sorted according to size so that the weight was between 160 to 210 g (average weight 188 g), corresponding to size 100 (100 fruit/bushel). The fruit were stored at 8 °C before being processed.

Internal ethylene concentration (IEC), color (both blush and shade sides), firmness, and titratable acidity of whole fruits were measured on the day of processing. Firmness was measured at 2 opposite sites on the equator of fruit after peel removal.

To measure IEC, apples were incubated overnight at ambient temperature (23 °C) before 0.5 mL gas from the core cavity were withdrawn through a septum attached to a needle. The gas was injected into a 5890 GC equipped with an 8M GS-GasPro capillary column (0.32 mm i.d. 0.25 µm film thickness) and a flame ionization detector (Agilent Technologies, Palo Alto, Calif., U.S.A.). Oven, injector, and detector temperatures were maintained at 50, 100, and 200 °C, respectively. The flow rates for carrier (He), make-up (He), hydrogen, and air were 1, 30, 30, and 360 mL/min, respectively. The detection limit for ethylene was approximately 1 ppm. Ethylene was calculated by comparing samples with a 10-ppm standard.

### Apple slice preparation and calcium ascorbate treatment

Processing of fresh-cut apples was performed in an 8 °C clear-processing room. All cutting boards, holding vessels, and then the fruit surfaces were sanitized with 200 ppm NaOCl (pH 9.2). An apple slicer was used to slice the apples into 8 equal pieces and to remove the core. The sliced apples were dropped into either water or 7% CaA solution (a concentration commonly used by the industry). All apple slices were treated between 2 to 3 min because the time required to slice 1 batch of apples was about 1 min. The slices were then drained and placed into CP08300 film bags (Cryovac, Duncan, S.C., U.S.A.) with O<sub>2</sub> and CO<sub>2</sub> transmission rates of 23.7 and 102.8 nmol/m<sup>2</sup>/s/kPa at 23 °C, respectively (4650 and 20150 mL/m<sup>2</sup>/24 h/atm at 23 °C, respectively). The bags were sealed using an AIE-300 heat sealer (American Intl. Electric, Whittier, Calif., U.S.A.). The bag sizes were approximately 14 × 16 cm, and there were 10 slices (approximately 150 g) per bag. The packaged apple slices were irradiated and then stored at 10 °C until analysis. Color, firmness, AA, TA, and pH were measured at 0, 1, 2, and 3 wk (1, 8, 15, and 22 d) of storage. O<sub>2</sub> and CO<sub>2</sub> in the package headspace were measured at 0, 1, 4, 8, 15, and 22 d.

### Irradiation and dosimetry

Apple slices in packages were either untreated (0 kGy) or treated with 0.5 or 1.0 kGy gamma radiation at a dose rate of 0.093 kGy/min at 7 ± 2 °C using a <sup>137</sup>Cs source. The custom-made, self-contained gamma radiation source (Lockheed Georgia Co., Marietta, Ga., U.S.A.) has 23 <sup>137</sup>Cs pencils placed in an annular array around a 63.5-cm high stainless steel cylindrical chamber with a 22.9-cm internal diameter.

Variations in radiation dose absorption were minimized by placing the samples within a uniform area of the radiation field, by irradiating them within a polypropylene container (4-mm wall) to absorb Compton electrons and by using the same geometry for samples during the entire study. Absorbed doses measured using alanine pellet dosimeters and a Bruker EMS 104 EPR analyzer were within 7% of targeted doses.

### Firmness measurement

Firmness was evaluated with a TA-XT2i Texture Analyzer (Texture Technologies Corp., Scarsdale, N.Y., U.S.A.). A 6-mm dia probe was used to penetrate peeled whole apples or the centers of the cutting surface of apple slices to 10 mm at 10 mm/s. Four slices from each bag were used for firmness measurements, and there were a total of 12 measurements for each treatment per experiment. Maximum force was recorded using the Texture Expert software (version 1.22, Texture Technologies Corp., Scarsdale, N.Y., U.S.A.).

### Color analysis

Color (CIE L\*, a\*, b\*) was measured with a ColorQuest XE colorimetric spectrophotometer (Hunter Associates Lab, Reston, Va., U.S.A.) using a 1-cm measuring aperture. The spectrophotometer was calibrated using the standard light trap and a white tile (L\* 93.50, a\* -0.89, and b\* 1.01). D65/10° were the illuminant/viewing geometry. Two readings were taken on each apple slice (one on each cutting surface) or the whole apple (blush and shade sides). Four apple slices and 4 whole apples were measured. Hue and chroma values were calculated from the following equations: Hue =  $\tan^{-1}(b^*/a^*)$  and chroma =  $[(a^*{}^2 + b^*{}^2)^{1/2}]$ .

### Determination of ascorbic acid

AA extraction and analysis were performed according to Graham and Annette (1992) with minor modifications. Samples (10 g) were homogenized with 20 mL of 5% metaphosphoric acid (MPA) using a Virtishear homogenizer (Virtis, Gardiner, N.Y., U.S.A.) at a speed setting of 70 for 1 min. The homogenate was filtered through a 4-layer cheesecloth, and then the filtrate was centrifuged at 12000 × g for 10 min at 4 °C in a Sorvall RC2-B refrigerated centrifuge (Kendro Laboratory Products, Newtown, Conn., U.S.A.). The supernatant was then used for the AA analysis. After proper dilutions, aliquots of supernatant were filtered through a 0.45 µm Acrodisc LC 13 PVDF syringe filter (Gelman Sciences, Ann Arbor, Mich., U.S.A.). The filtered samples were placed into 2-mL vials and analyzed using a Hewlett Packard Ti-series 1050 high-performance liquid chromatography system (Agilent Technologies, Palo Alto, Calif., U.S.A.). The injection volume was 20 µL. Separation of compounds was achieved with an Aminex HPX-87H organic acids column (300 H 7.8 mm) fitted with a microguard cation H<sup>+</sup> eluted with a mobile phase of 5 mM sulfuric acid at a flow rate of 0.5 mL/min. The column temperature was maintained at 30 °C using a column heater (Bio-Rad Laboratories, Hercules, Calif., U.S.A.). AA was monitored at 245 nm and calculated from an AA standard.

### Headspace gas analysis

Headspace gases within the sealed packages were sampled to determine the levels of CO<sub>2</sub> and O<sub>2</sub> during storage. To measure the atmosphere, a 0.5-mL headspace sample was withdrawn from the bags by piercing the film with a fine hypodermic needle on a syringe. The sampling hole was resealed with electrical tape, and the contents of the bag were used in subsequent analysis. The gas samples were then injected into a Gow-Mac Series 580 gas chromatograph (Gow-Mac Instrument, Bridgewater, N.J., U.S.A.) equipped with a 183 cm CTR I column (Alltech Associates, Inc., Deerfield, Ill., U.S.A.) and a thermal conductivity detector. The CTR I column consists of an outer

column (0.64 cm i.d.) packed with an activated molecular sieve and an inner column (0.32 cm i.d.) packed with a porous polymer mixture. The injector, oven, and detector temperatures were held at ambient temperature (23 °C). The carrier gas was helium with a flow rate of 120 mL/min. A measurement was made from each bag, and there were a total of 6 measurements for each treatment. CO<sub>2</sub> and O<sub>2</sub> levels were calculated in comparison to a standard.

### Analysis of pH and titratable acidity

Juice was extracted from apple slices using a Champion MAR-48C juicer (Plastaket MFG Co., Lodi, Calif., U.S.A.). The juice was stored at -20 °C until analysis. The pH was recorded before titration. The TA was measured by titrating a 5-mL aliquot of juice to pH 8.1 using an autotitrator (Radiometer Analytical, Lyon, France).

### Microflora

The microflora were measured at 0, 1, 5, 8, 15, and 22 d of storage. Each sample consisted of a stomacher bag containing apples slices, typically 50 to 60 g of material per bag. The precise weight of material was recorded for each sample, and sterile Butterfield's phosphate buffer (BPB) equal to 4 times sample weight was added, e.g., 54 g apple + 216 mL BPB. The sample bags were closed and palpitated (60 s) to obtain a surface wash of the sample. A 1-mL aliquot of the wash buffer was withdrawn and serially diluted in BPB in 1:10 increments to a final dilution of 10<sup>5</sup>. Separate 1 mL aliquots of each of the dilutions from 10<sup>0</sup> to 10<sup>5</sup> were withdrawn and pour plated with tryptic soy agar (TSA), 3 plates per dilution. The plates were allowed to cool, inverted, and incubated at 37 °C overnight. The plates were counted with a calibrated AccuCount 1000 automated colony counter (Biologics, Gainesville, Va., U.S.A.). The plate count values obtained, representing colony-forming units (CFU)/mL of wash buffer, were back-calculated to account for dilution and weight of tissue to provide the final CFU/g tissue values presented in Figure 2.

### Experimental design and statistical analysis

The experimental design was a split-plot design with the whole plot factor being the CaA treatment and the subplots consisting of completely randomized design of the radiation doses and storage times. Two trials were performed 3 d apart. Within each trial, slices prepared from each of 3 batches of apples were subjected to each

**Table 1—Maturity and quality parameters (internal ethylene concentration [IEC], firmness, titratable acidity [TA], pH, background and flush side hue values) of 'Gala' apples used on the day for the preparation of apple slices<sup>a,b</sup>**

IEC (ppm)	Firmness (kg)	TA (%)	pH	Hue (background)	Hue (flush)
Exp no 1					
0.0 ± 0.0	2.3 ± 0.4	0.267 ± 0.032	4.1 ± 0.3	73.9 ± 15.5	25.4 ± 6.7
Exo no 2					
0.4 ± 1.2	2.4 ± 0.4	0.282 ± 0.049	4.0 ± 0.3	82.7 ± 7.5	26.1 ± 6.5

<sup>a</sup>Values are means ± standard deviations, *n* = 12.

<sup>b</sup>There was no significant (*P* > 0.05) difference between experiment 1 and 2 for any parameter.

treatment (all possible combination of CaA treatment and irradiation doses). The data from the 3 batches collected on the 1st trial were combined with the data from the 3 batches collected on the 2nd trial to give 6 replicates. The data were analyzed with SAS version 7 (SAS Inst., Cary, N.C., U.S.A.). Effects of storage time, radiation dose, and CaA were analyzed using the least significant difference (LSD) test of General Linear Models procedure. Only significant (*P* < 0.05) results are discussed unless stated otherwise.

## Results and Discussion

### Whole fruit maturity and quality

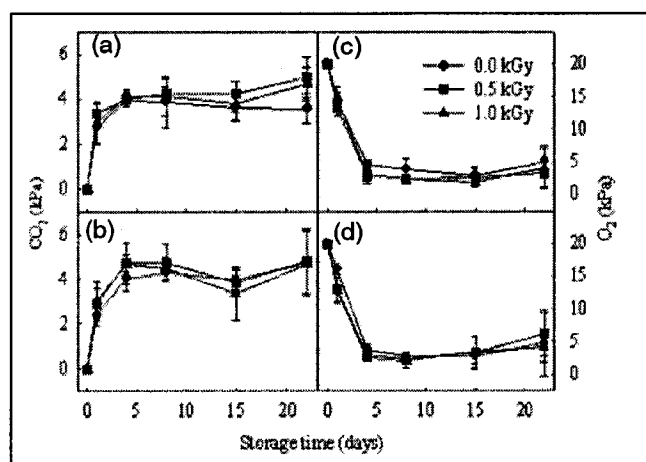
The maturity and quality parameters of fruit used in the 2 experiments (trials) are listed in Table 1. The IEC of all fruit tested was below 1 ppm except for 1 fruit in the 2nd experiment with 5.4 ppm, indicating the fruit used in the study were mostly in the pre-climacteric stage. Firmness, TA, pH, and hue of the fruit were not different between the 2 experiments, suggesting that the fruit for the 2 experiments had similar maturity. The data for the sliced apples were therefore combined.

### Headspace atmosphere

The O<sub>2</sub> and CO<sub>2</sub> levels in packages are presented in Figure 1. The CO<sub>2</sub> levels in all packages, regardless of CaA treatment or radiation dose, increased rapidly, reaching 4 to 5 kPa (1 kPa 1%) at day 4 and stayed relatively constant during the rest of the storage period. The O<sub>2</sub> levels decreased rapidly to 3 to 4 kPa (%) during the 1st 4 d and maintained these levels during the rest of the storage period. CaA treatment and irradiation had no effect on headspace composition. The increase in CO<sub>2</sub> and reduction in O<sub>2</sub> in the packages are presumably due to respiration of apple slices. Earlier studies have shown that irradiation increased respiration rate in fresh-cut apples (Gunes and others 2000). Our results showed that although CO<sub>2</sub> levels were higher and O<sub>2</sub> levels were lower in irradiated packages overall, the O<sub>2</sub> and CO<sub>2</sub> levels in irradiated and nonirradiated packages were not significantly (*P* > 0.05) different on any given day. This suggested the effect of low dose radiation on respiration did not substantially alter gas composition in MAP in our test. Rocha and Moraes (2001) found that high CO<sub>2</sub> and low O<sub>2</sub> levels retain firmness and extend shelf-life of cut apples by inhibiting polyphenol oxidase activity. The optimum O<sub>2</sub> level for fresh-cut apples is probably similar to the level for whole apples, which is around 1 kPa (%) (Gorny 1997). Therefore, the O<sub>2</sub> levels in our study were higher than the optimum level. The similarity of gas composition in irradiated and nonirradiated packages suggests any effect of CaA and irradiation on quality is not due to headspace atmosphere.

### Firmness

Firmness was affected by CaA treatment, radiation dose, and storage time (Table 2). At wk 0, firmness of fresh-cut apples decreased as



**Figure 1—Changes in CO<sub>2</sub> (a and b) and O<sub>2</sub> (c and d) levels measured at 0, 1, 4, 8, 15, and 22 d of storage for the headspace of film bags that contained fresh-cut apple slices treated with (a, c) or without (b, d) 7% calcium ascorbate and irradiated with 0-, 0.5-, and 1.0-kGy gamma radiation. There was no significant (*P* > 0.05) effect of irradiation dose on CO<sub>2</sub> and O<sub>2</sub> levels on any given day.**

**Table 2—Effects of irradiation dose and calcium ascorbate on firmness (kg) of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
<b>0% calcium ascorbate</b>				
0	2.33 ± 0.36	2.07 ± 0.45	1.80 ± 0.27	0.21
1	2.02 ± 0.56	1.65 ± 0.39	1.52 ± 0.31	0.25
2	1.85 ± 0.35	1.68 ± 0.36	1.47 ± 0.34	0.20
3	1.85 ± 0.43	1.49 ± 0.34	1.41 ± 0.32	0.21
LSD <sup>b</sup>	0.25	0.22	0.18	
<b>7% calcium ascorbate</b>				
0	2.41 ± 0.30	2.10 ± 0.36	1.90 ± 0.29	0.18
1	2.38 ± 0.39	1.99 ± 0.48	1.80 ± 0.31	0.23
2	2.02 ± 0.47	1.87 ± 0.41	1.71 ± 0.33	0.23
3	2.04 ± 0.47	1.97 ± 0.31	1.70 ± 0.32	0.21
LSD <sup>b</sup>	0.24	0.23	0.18	
CaA effect	**c	***	***	

<sup>a</sup>Values are means ± standard deviations, *n* = 6.<sup>b</sup>The least significant difference (LSD) at *P* < 0.05 level.<sup>c</sup>\*\* and \*\*\* indicate significant difference at *P* < 0.01 and *P* < 0.001 levels, respectively, for the CaA effect.

radiation dose increased, regardless of CaA treatment. Treatment with CaA did not reduce the loss due to irradiation. The decrease in firmness was approximately 22% per kGy. Firmness of apple slices of all treatments decreased during storage except for slices treated with 7% CaA at 0.5 kGy radiation. However, the loss of firmness due to storage was reduced by CaA treatment. During the 3 wk of storage, the loss of firmness of apple slices without CaA treatment was 24% while the loss in CaA treated slices was only 11%. Because CaA treatment reduced firmness loss during storage, fresh-cut apples treated with CaA had a higher firmness than those without CaA treatment after 1, 2, and 3 wk of storage. After 2 to 3 wk of storage, apple slices treated with 7% CaA and 0.5 kGy radiation had similar or higher firmness than the nonirradiated samples that were not treated with CaA.

### The pH and titratable acidity

The pH of apple slices treated with CaA was slightly (but not always significantly) higher than the non-treated samples (data not shown). Irradiation and storage had no consistent effect on pH.

The effects of irradiation dose, CaA, and storage time on TA are presented in Table 3. Irradiation had no significant effect on TA in the sliced apples. Fan and Mattheis (2001) found that irradiation decreased TA in whole apples. TA tended to decrease during the 1st wk of storage in all treatments except the nonirradiated slices with CaA treatment. However, there were no significant decreases during the remaining part of the storage period compared with wk 1. During storage, fresh-cut apples respire and utilize organic acids quickly compared with other compounds, resulting in a decreased TA (Moller and Palmer 1984). This decrease during the 1st wk of storage may be due to increased respiration following tissue damage (cutting).

### Ascorbic acid content

The initial AA levels in CaA-treated apple slices were approximately 70 times higher than the nontreated ones (Table 4), indicating that ascorbate penetrated into fruit tissue (and/or attached to slice surfaces). AA levels decreased rapidly during storage, particularly during the 1st week. Nearly half of AA was lost in samples without CaA treatment during the 1st week, but during the rest of the storage period, AA levels were maintained. In the apple slices treated with CaA, AA content decreased continuously during 3 wk of storage, though more loss was observed in the 1st wk in terms of absolute amount. Despite a higher rate of AA loss in the CaA-treated apple slices, the AA levels of CaA-treated apple slices were

**Table 3—Effects of irradiation dose and calcium ascorbate on titratable acidity (%) of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
0% calcium ascorbate				
0	0.258 ± 0.023	0.273 ± 0.030	0.244 ± 0.024	0.032
1	0.225 ± 0.028	0.224 ± 0.032	0.206 ± 0.022	0.038
2	0.235 ± 0.021	0.234 ± 0.018	0.230 ± 0.009	0.021
3	0.233 ± 0.053	0.231 ± 0.028	0.233 ± 0.027	0.047
LSD <sup>b</sup>	0.041	0.033	0.027	
7% calcium ascorbate				
0	0.227 ± 0.021	0.240 ± 0.014	0.232 ± 0.019	0.023
1	0.231 ± 0.025	0.205 ± 0.033	0.221 ± 0.036	0.039
2	0.220 ± 0.031	0.212 ± 0.012	0.229 ± 0.028	0.031
3	0.230 ± 0.013	0.234 ± 0.023	0.210 ± 0.033	0.030
LSD <sup>b</sup>	0.029	0.027	0.036	
CaA effect	NS <sup>c</sup>	*	NS	

<sup>a</sup>Values are means ± standard deviations, *n* = 6.<sup>b</sup>The least significant difference (LSD) at *P* < 0.05 level.<sup>c</sup>NS and \* indicate not significant and significant difference at *P* < 0.05 level, respectively, for the CaA effect.**Table 4—Effects of irradiation dose and calcium ascorbate on ascorbic acid contents (mg/100 g fresh weight) of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
<b>0% calcium ascorbate</b>				
0	2.1 ± 0.9	1.8 ± 0.7	1.9 ± 0.9	1.0
1	1.3 ± 0.8	1.0 ± 0.3	0.9 ± 0.2	0.6
2	1.4 ± 0.6	0.9 ± 0.2	0.9 ± 0.3	0.5
3	1.2 ± 0.6	1.2 ± 0.4	1.0 ± 0.4	0.6
LSD <sup>b</sup>	0.9	0.6	0.6	
<b>7% calcium ascorbate</b>				
0	139.9 ± 19.1	127.9 ± 24.6	132.3 ± 21.5	20.9
1	83.2 ± 28.7	83.4 ± 21.9	86.2 ± 15.9	28.1
2	57.2 ± 14.6	59.3 ± 14.5	56.2 ± 15.6	18.4
3	38.0 ± 12.0	34.5 ± 19.3	45.8 ± 7.6	16.8
LSD <sup>b</sup>	23.6	24.6	19.2	
CaA effect	****	***	***	

<sup>a</sup>Values are means ± standard deviations, *n* = 6.<sup>b</sup>The least significant difference (LSD) at *P* < 0.05 level.<sup>c</sup>\*\*\* indicates significant difference at *P* < 0.001 level for the CaA effect.

still about 30 to 45 times higher than the nontreated slices after 3 wk of storage. Irradiation at doses of 0.5 and 1.0 kGy had no significant (*P* > 0.05) effect on AA levels of either CaA treated or nontreated apples and did not affect the AA loss during storage on any given day. CaA treatment should provide more nutritious apples slices by adding AA and Ca (Chardonnet and others 2002).

### Color changes

The *L*\* values (lightness) of slices increased in all treatments during the 1st wk of storage (Table 5). During the rest of storage, the *L*\* values tended to decrease. Except for the nonirradiated samples treated with CaA, the *L*\* values decreased from wk 2 to wk 3 for all treatments. Irradiation at both doses caused slight but significant (*P* < 0.05) decreases in *L*\* values of slices treated with or without CaA at wk 0, and in those without CaA at wk 2 and 3, even though the effect may not be visually noticeable. Overall, CaA treatment increased *L*\* values of irradiated slices, indicating the CaA-treated samples were whiter.

The hue value represents true color, and the hue values for the apple slices are presented in Table 6. In this study, as slices turn brown, the hue values decrease. The hue values of apple slices of all treatments decreased during storage, indicating surface browning

**Table 5—Effects of irradiation dose and calcium ascorbate on lightness ( $L^*$ ) of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
<b>0% calcium ascorbate</b>				
0	72.9 ± 2.2	71.8 ± 1.6	71.5 ± 1.7	0.7
1	78.1 ± 2.6	78.4 ± 1.9	78.1 ± 2.0	0.9
2	77.8 ± 2.1	75.5 ± 3.6	76.7 ± 2.2	1.1
3	76.0 ± 2.3	70.4 ± 5.7	69.4 ± 8.6	2.5
LSD <sup>b</sup>	0.9	1.4	1.9	
<b>7% calcium ascorbate</b>				
0	74.1 ± 1.3	73.0 ± 1.6	72.9 ± 2.0	0.7
1	79.4 ± 1.5	79.0 ± 1.8	79.9 ± 1.5	0.6
2	76.6 ± 6.3	78.1 ± 2.6	78.2 ± 2.0	1.7
3	76.6 ± 2.6	76.0 ± 3.3	74.0 ± 5.1	1.5
LSD <sup>b</sup>	1.4	1.0	1.2	
CaA effect	NS <sup>c</sup>	***	***	

<sup>a</sup>Values are means ± standard deviations,  $n = 6$ .<sup>b</sup>The least significant difference (LSD) at  $P < 0.05$  level.<sup>c</sup>NS and \*\*\* indicate not significant and significant at  $P < 0.001$  level, respectively, for the CaA effect.

occurred during storage. A slight but significant ( $P < 0.05$ ) decrease in hue values at a given storage time due to irradiation was usually observed. Samples that were treated with CaA always had higher hue values than those without CaA regardless if irradiation was applied, indicating that CaA is an effective anti-browning inhibitor even after irradiation. Because irradiation did not have a significant effect on AA levels in apple slices (Table 4), AA was still effective in inhibiting enzymatic browning in irradiated apple slices.

Chroma describes the saturation of a color and is presented in Table 7 for apple slices in the present study. CaA treatment reduced chroma values of both irradiated and nonirradiated samples measured at wk 0. These changes in chroma were mainly due to decreases in  $b^*$  values (blueness-yellow chromatism), indicating the CaA-treated samples became less yellow. However, the loss of yellowness in the apples slices was reversible because the  $b^*$  and chroma of the CaA-treated samples became more similar to the nontreated samples after the 1st wk of storage. Chroma values increased in apple slices treated with CaA while chroma values in samples without CaA either did not increase or increased to a lesser degree during the rest of storage period. Irradiation decreased chroma values of CaA-treated samples at wk 1, 2, and 3, however, no irradiation-induced decrease in chroma values was observed in samples without CaA. Changes in  $a^*$  and  $L^*$  values have been used in monitoring enzymatic browning on fresh-cut apple surfaces (Soliva-Fortuny and others 2001). The  $L^*$  and  $a^*$  values were correlated with polyphenol oxidase activity, but no correlation was observed for  $b^*$  and chroma values with polyphenol oxidase activity (Kim and others 1995), suggesting that changes in  $b^*$  and chroma may not be related to enzymatic browning. The mechanisms on the CaA-induced decrease in  $b^*$  and chroma values and their recoverability are unknown. The CaA may induce decoloration of apple pigments (that is, tissue bleaching) evidenced by the visually observed whitening of the cut apple surface after CaA treatment.

### Microflora

The microflora counts as functions of CaA and irradiation treatments with storage are shown in Figure 2. CaA had no significant effect on microflora population at any given storage time and radiation level. The initial total aerobic plate count (TAPC) of all samples was near or below the detection limit. The TAPC of the samples was much lower than many vegetable samples (Fan and others 2003). During storage, TAPC increased in all samples. At wk 1, TAPC of nonirradiated samples was approximately 3.5 log CFU/g. TAPC for

**Table 6—Effects of irradiation dose and calcium ascorbate on hue values of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
<b>0% calcium ascorbate</b>				
0	87.7 ± 1.7	87.0 ± 1.5	86.9 ± 1.6	0.7
1	86.7 ± 1.7	85.5 ± 2.0	84.9 ± 2.0	0.8
2	86.2 ± 1.4	83.9 ± 2.5	84.8 ± 1.6	0.8
3	85.2 ± 2.0	82.5 ± 2.9	81.0 ± 4.5	1.3
LSD	0.7	0.9	1.1	
<b>7% calcium ascorbate</b>				
0	94.6 ± 2.2	93.8 ± 2.4	93.4 ± 1.9	0.9
1	94.5 ± 1.4	93.0 ± 2.2	93.3 ± 1.6	0.7
2	92.3 ± 7.5	92.8 ± 2.2	92.8 ± 2.0	1.9
3	92.8 ± 2.5	91.3 ± 2.4	90.2 ± 2.9	1.1
LSD	1.7	0.9	0.9	
CaA effect	*** <sup>c</sup>	***	***	

<sup>a</sup>Values are means ± standard deviations,  $n = 6$ .<sup>b</sup>The least significant difference (LSD) at  $P < 0.05$  level.<sup>c</sup>\*\*\* indicates significant difference at  $P < 0.001$  level for the CaA effect.**Table 7—Effects of irradiation dose and calcium ascorbate on chroma of apple slices during storage at 10 °C<sup>a</sup>**

Storage time (wk)	Radiation dose (kGy)			LSD <sup>b</sup>
	0	0.5	1.0	
<b>0% calcium ascorbate</b>				
0	19.0 ± 2.3	19.4 ± 2.4	19.7 ± 1.9	0.9
1	20.0 ± 2.4	19.5 ± 2.3	19.7 ± 2.4	1.0
2	19.0 ± 2.0	20.6 ± 2.0	20.0 ± 2.3	0.9
3	18.9 ± 2.0	20.8 ± 2.9	20.2 ± 3.2	1.1
LSD <sup>b</sup>	0.9	1.0	1.0	
<b>7% calcium ascorbate</b>				
0	15.1 ± 2.3	15.6 ± 2.3	15.8 ± 2.1	0.9
1	20.4 ± 1.9	18.6 ± 2.0	17.6 ± 1.5	0.7
2	21.1 ± 3.9	19.1 ± 2.5	18.7 ± 2.1	1.2
3	21.4 ± 2.8	20.3 ± 2.8	19.4 ± 2.1	1.0
LSD <sup>b</sup>	1.1	1.0	0.8	
CaA effect	NS <sup>c</sup>	*** <sup>c</sup>	*** <sup>c</sup>	

<sup>a</sup>Values are means ± standard deviations,  $n = 6$ .<sup>b</sup>The least significant difference (LSD) at  $P < 0.05$  level.<sup>c</sup>NS and \*\*\* indicate not significant and significant difference at  $P < 0.001$  levels, respectively, for the CaA effect.

the 0.5-kGy samples was in the range of 2.1 to 2.4 log CFU while 1.0 kGy radiation further reduced the TAPC to approximately 1 log CFU/g. Similar reduction in TAPC by irradiation was observed at wk 2. At wk 3, TAPC of nonirradiated samples and those samples that received 0.5 kGy radiation became similar while 1.0 kGy samples had a roughly 2 log lower TAPC than nonirradiated samples. Radiation at 1 kGy significantly reduced microflora population throughout the 3-wk storage period, but the significant reduction in TAPC due to the 0.5-kGy radiation was only observed for the 1st 2 wk of storage. This study further demonstrated that low dose ionizing radiation can usually improve microbiological quality of sliced apples.

We used 10 °C to mimic the possible temperature abuse that occurs during transportation, storage, and display. Microorganisms regrew during storage at the abused temperature. The subsequent growth of microorganisms will be reduced if fresh-cut apples are stored at their optimum storage temperature of 0 to 5 °C (Gorny 1997) instead of 10 °C as used in this experiment. Future research is needed to study the growth of food-borne pathogens after irradiation of fresh-cut apple slices under different storage temperatures.

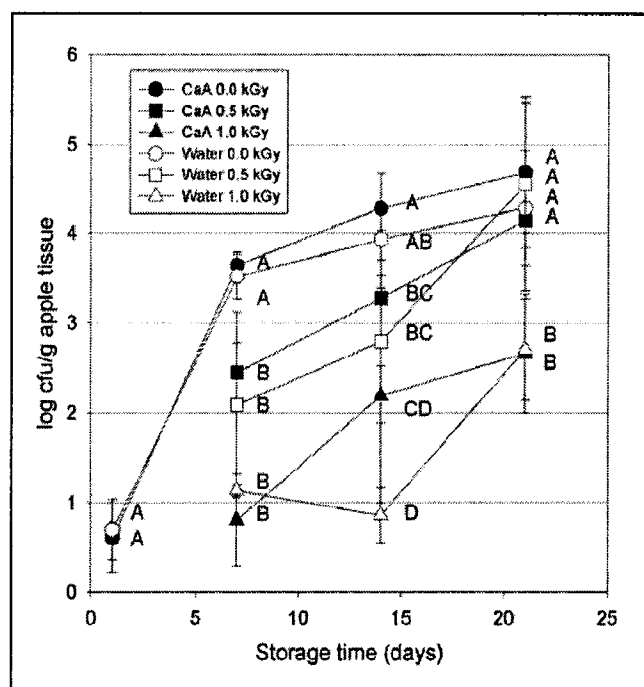
The shelf life of fresh-cut apples stored at 10 °C cannot be extended to 3 wk as shown by browning, a soggy appearance, and growth of microorganisms. The appearance of apple slices was

more desirable for the CaA-treated apples than those without CaA treatment at wk 3. The  $L^*$  values of irradiated samples, particularly those without CaA, decreased rapidly during the last wk of the storage, and this darkening was readily observed. It is unclear what causes the decrease in  $L^*$  values in the irradiated samples. The changes may be related to radiation-induced synthesis of phenolic compounds and tissue browning.

CaA did not directly influence the radiation effect on firmness but did reduce the firmness loss of irradiated apple slices during storage. Rocha and others (1998) also found AA reduced firmness loss of fresh-cut apples during storage. Irradiation-induced softening may be due to the breakdown of cellular constituents such as pectin, cellulose and hemicellulose, and alteration of cell membrane, resulting in structural weakening and loss of turgor (Poovaiah and others 1988). Kovacs and others (1988) suggested that the mechanism by which calcium maintained firmness in irradiated whole apples was more related to the stabilization of cell membranes by calcium than to cross-linking of pectic acids by calcium to form calcium pectate in cell walls. The reduced rate of firmness loss in apple slices containing CaA may be mostly due to the role of calcium in cell membranes and the antioxidant activity of ascorbate.

### Conclusions

Irradiation had little effect on AA content, TA, or pH of fresh-cut fruits, reduced fruit firmness, and the population of microflora, but slightly increased browning. Use of CaA dipping inhibited surface browning, reduced the firmness loss during storage, and increased AA content in fruit tissues. Therefore, the firmness loss and browning in fresh-cut apple slices resulting from irradiation can be compensated by the CaA dipping. A combination of CaA dipping and low dose ionizing radiation resulted in a microbiologically safe and high quality of fresh-cut apples rich in nutrients (CaA and AA).



**Figure 2—Total aerobic plate count (TAPC) of fresh-cut apples during storage at 10 °C. All data points represent means from 6 analyses accompanied by standard deviations. Samples that received 0.5- and 1.0-kGy radiation had TAPC below the detection limit at day 1. Means of TAPC not containing a common letter at a given storage time are significantly ( $P < 0.05$ ) different from each other by the LSD test.**

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